



SEQUENCE LISTING

<110> Bron, Sierd
Jongbloed, Jan D.H.
Mueller, Joerg P.
Van Dijl, Jan M.

<120> Twin-Arginine Translocation in Bacillus

<130> GC634-2

<140> US 09/954,737

<141> 2001-09-17

<150> US 60/233,610

<151> 2000-09-18

<160> 29

<170> FastSEQ for Windows Version 4.0

<210> 1

<211> 89

<212> PRT

<213> Escherichia coli

<400> 1

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Gly | Gly | Ile | Ser | Ile | Trp | Gln | Leu | Leu | Ile | Ile | Ala | Val | Ile | Val |
| 1 | | | | 5 | | | | | 10 | | | | | 15 | |
| Val | Leu | Leu | Phe | Gly | Thr | Lys | Lys | Leu | Gly | Ser | Ile | Gly | Ser | Asp | Leu |
| | | | 20 | | | | | 25 | | | | | 30 | | |
| Gly | Ala | Ser | Ile | Lys | Gly | Phe | Lys | Lys | Ala | Met | Ser | Asp | Asp | Glu | Pro |
| | | 35 | | | | | 40 | | | | | 45 | | | |
| Lys | Gln | Asp | Lys | Thr | Ser | Gln | Asp | Ala | Asp | Phe | Thr | Ala | Lys | Thr | Ile |
| | 50 | | | | | 55 | | | | 60 | | | | | |
| Ala | Asp | Lys | Gln | Ala | Asp | Thr | Asn | Gln | Glu | Gln | Ala | Lys | Thr | Glu | Asp |
| 65 | | | | | 70 | | | | 75 | | | | | 80 | |
| Ala | Lys | Arg | His | Asp | Lys | Glu | Gln | Val | | | | | | | |
| | | | | 85 | | | | | | | | | | | |

<210> 2

<211> 67

<212> PRT

<213> Escherichia coli

<400> 2

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Gly | Glu | Ile | Ser | Ile | Thr | Lys | Leu | Leu | Val | Val | Ala | Ala | Leu | Val |
| 1 | | | | 5 | | | | | 10 | | | | | 15 | |
| Val | Leu | Leu | Phe | Gly | Thr | Lys | Lys | Leu | Arg | Thr | Leu | Gly | Gly | Asp | Leu |
| | | | 20 | | | | | 25 | | | | | 30 | | |
| Gly | Ala | Ala | Ile | Lys | Gly | Phe | Lys | Lys | Ala | Met | Asn | Asp | Asp | Asp | Ala |
| | | 35 | | | | | 40 | | | | | 45 | | | |
| Ala | Ala | Lys | Lys | Gly | Ala | Asp | Val | Asp | Leu | Gln | Ala | Glu | Lys | Leu | Ser |
| | 50 | | | | | 55 | | | | 60 | | | | | |
| His | Lys | Glu | | | | | | | | | | | | | |

65

<210> 3
<211> 57
<212> PRT
<213> *Bacillus subtilis*

<400> 3
Met Pro Ile Gly Pro Gly Ser Leu Ala Val Ile Ala Ile Val Ala Leu
1 5 10 15
Ile Ile Phe Gly Pro Lys Lys Leu Pro Glu Leu Gly Lys Ala Ala Gly
20 25 30
Asp Thr Leu Arg Glu Phe Lys Asn Ala Thr Lys Gly Leu Thr Ser Asp
35 40 45
Glu Glu Glu Lys Lys Lys Glu Asp Gln
50 55

<210> 4
<211> 70
<212> PRT
<213> *Bacillus subtilis*

<400> 4
Met Phe Ser Asn Ile Gly Ile Pro Gly Leu Ile Leu Ile Phe Val Ile
1 5 10 15
Ala Ile Ile Ile Phe Gly Pro Ser Lys Leu Pro Glu Ile Gly Arg Ala
20 25 30
Ala Lys Arg Thr Leu Leu Glu Phe Lys Ser Ala Thr Lys Ser Leu Val
35 40 45
Ser Gly Asp Glu Lys Glu Glu Lys Ser Ala Glu Leu Thr Ala Val Lys
50 55 60
Gln Asp Lys Asn Ala Gly
65 70

<210> 5
<211> 62
<212> PRT
<213> *Bacillus subtilis*

<400> 5
Met Glu Leu Ser Phe Thr Lys Ile Leu Val Ile Leu Phe Val Gly Phe
1 5 10 15
Leu Val Phe Gly Pro Asp Lys Leu Pro Ala Leu Gly Arg Ala Ala Gly
20 25 30
Lys Ala Leu Ser Glu Phe Lys Gln Ala Thr Ser Gly Leu Thr Gln Asp
35 40 45
Ile Arg Lys Asn Asp Ser Glu Asn Lys Glu Asp Lys Gln Met
50 55 60

<210> 6
<211> 171
<212> PRT
<213> *Escherichia coli*

<400> 6
Met Phe Asp Ile Gly Phe Ser Glu Leu Leu Val Phe Ile Ile Gly
1 5 10 15

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Leu | Val | Val | Leu | Gly | Pro | Gln | Arg | Leu | Pro | Val | Ala | Val | Lys | Thr | Val |
| | | | 20 | | | | | 25 | | | | | 30 | | |
| Ala | Gly | Trp | Ile | Arg | Ala | Leu | Arg | Ser | Leu | Ala | Thr | Thr | Val | Gln | Asn |
| | | 35 | | | | | 40 | | | | | 45 | | | |
| Glu | Leu | Thr | Gln | Glu | Leu | Lys | Leu | Gln | Glu | Phe | Gln | Asp | Ser | Leu | Lys |
| | 50 | | | | | 55 | | | | | 60 | | | | |
| Lys | Val | Glu | Lys | Ala | Ser | Leu | Thr | Asn | Leu | Thr | Pro | Glu | Leu | Lys | Ala |
| | 65 | | | | 70 | | | | | 75 | | | | | 80 |
| Ser | Met | Asp | Glu | Leu | Arg | Gln | Ala | Ala | Glu | Ser | Met | Lys | Arg | Ser | Tyr |
| | | | | 85 | | | | | 90 | | | | | 95 | |
| Val | Ala | Asn | Asp | Pro | Glu | Lys | Ala | Ser | Asp | Glu | Ala | His | Thr | Ile | His |
| | | | 100 | | | | | | 105 | | | | 110 | | |
| Asn | Pro | Val | Val | Lys | Asp | Asn | Glu | Ala | Ala | His | Glu | Gly | Val | Thr | Pro |
| | | 115 | | | | | 120 | | | | | 125 | | | |
| Ala | Ala | Ala | Gln | Thr | Gln | Ala | Ser | Ser | Pro | Glu | Gln | Lys | Pro | Glu | Thr |
| | | 130 | | | | 135 | | | | | 140 | | | | |
| Thr | Pro | Glu | Pro | Val | Val | Lys | Pro | Ala | Ala | Asp | Ala | Glu | Pro | Lys | Thr |
| | | | | | 150 | | | | | 155 | | | | | 160 |
| Ala | Ala | Pro | Ser | Pro | Ser | Ser | Ser | Asp | Lys | Pro | | | | | |
| | | | | 165 | | | | | 170 | | | | | | |

<210> 7

<211> 258

<212> PRT

<213> Escherichia coli

<400> 7

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Met | Ser | Val | Glu | Asp | Thr | Gln | Pro | Leu | Ile | Thr | His | Leu | Ile | Glu | Leu |
| 1 | | | | 5 | | | | | 10 | | | | | 15 | |
| Arg | Lys | Arg | Leu | Leu | Asn | Cys | Ile | Ile | Ala | Val | Ile | Val | Ile | Phe | Leu |
| | | | 20 | | | | | 25 | | | | | 30 | | |
| Cys | Leu | Val | Tyr | Phe | Ala | Asn | Asp | Ile | Tyr | His | Leu | Val | Ser | Ala | Pro |
| | | 35 | | | | | 40 | | | | | 45 | | | |
| Leu | Ile | Lys | Gln | Leu | Pro | Gln | Gly | Ser | Thr | Met | Ile | Ala | Thr | Asp | Val |
| | 50 | | | | | 55 | | | | | 60 | | | | |
| Ala | Ser | Pro | Phe | Phe | Thr | Pro | Ile | Lys | Leu | Thr | Phe | Met | Val | Ser | Leu |
| | 65 | | | | 70 | | | | | 75 | | | | | 80 |
| Ile | Leu | Ser | Ala | Pro | Val | Ile | Leu | Tyr | Gln | Val | Trp | Ala | Phe | Ile | Ala |
| | | | | 85 | | | | | 90 | | | | | 95 | |
| Pro | Ala | Leu | Tyr | Lys | His | Glu | Arg | Arg | Leu | Val | Val | Pro | Leu | Leu | Val |
| | | 100 | | | | | | 105 | | | | | 110 | | |
| Ser | Ser | Ser | Leu | Leu | Phe | Tyr | Ile | Gly | Met | Ala | Phe | Ala | Tyr | Phe | Val |
| | | 115 | | | | | 120 | | | | | 125 | | | |
| Val | Phe | Pro | Leu | Ala | Phe | Gly | Phe | Leu | Ala | Asn | Thr | Ala | Pro | Glu | Gly |
| | | 130 | | | | 135 | | | | | 140 | | | | |
| Val | Gln | Val | Ser | Thr | Asp | Ile | Ala | Ser | Tyr | Leu | Ser | Phe | Val | Met | Ala |
| | 145 | | | | 150 | | | | | 155 | | | | | 160 |
| Leu | Phe | Met | Ala | Phe | Gly | Val | Ser | Phe | Glu | Val | Pro | Val | Ala | Ile | Val |
| | | | | 165 | | | | | 170 | | | | | 175 | |
| Leu | Leu | Cys | Trp | Met | Gly | Ile | Thr | Ser | Pro | Glu | Asp | Leu | Arg | Lys | Lys |
| | | 180 | | | | | | 185 | | | | | 190 | | |
| Arg | Pro | Tyr | Val | Leu | Val | Gly | Ala | Phe | Val | Val | Gly | Met | Leu | Leu | Thr |
| | | 195 | | | | | 200 | | | | | 205 | | | |
| Pro | Pro | Asp | Val | Phe | Ser | Gln | Thr | Leu | Leu | Ala | Ile | Pro | Met | Tyr | Cys |
| | | 210 | | | | 215 | | | | | 220 | | | | |
| Leu | Phe | Glu | Ile | Gly | Val | Phe | Phe | Ser | Arg | Phe | Tyr | Val | Gly | Lys | Gly |
| | 225 | | | | 230 | | | | | 235 | | | | | 240 |

Arg Asn Arg Glu Glu Glu Asn Asp Ala Glu Ala Glu Ser Glu Lys Thr
245 250 255
Glu Glu

<210> 8
<211> 254
<212> PRT
<213> Bacillus subtilis

<400> 8
Met Thr Arg Met Lys Val Asn Gln Met Ser Leu Leu Glu His Ile Ala
1 5 10 15
Glu Leu Arg Lys Arg Leu Leu Ile Val Ala Leu Ala Phe Val Val Phe
20 25 30
Phe Ile Ala Gly Phe Phe Leu Ala Lys Pro Ile Ile Val Tyr Leu Gln
35 40 45
Glu Thr Asp Glu Ala Lys Gln Leu Thr Leu Asn Ala Phe Asn Leu Thr
50 55 60
Asp Pro Leu Tyr Val Phe Met Gln Phe Ala Phe Ile Ile Gly Ile Val
65 70 75 80
Leu Thr Ser Pro Val Ile Leu Tyr Gln Leu Trp Ala Phe Val Ser Pro
85 90 95
Gly Leu Tyr Glu Lys Glu Arg Lys Val Thr Leu Ser Tyr Ile Pro Val
100 105 110
Ser Ile Leu Leu Phe Leu Ala Gly Leu Ser Phe Ser Tyr Tyr Ile Leu
115 120 125
Phe Pro Phe Val Val Asp Phe Met Lys Arg Ile Ser Gln Asp Leu Asn
130 135 140
Val Asn Gln Val Ile Gly Ile Asn Glu Tyr Phe His Phe Leu Leu Gln
145 150 155 160
Leu Thr Ile Pro Phe Gly Leu Leu Phe Gln Met Pro Val Ile Leu Met
165 170 175
Phe Leu Thr Arg Leu Gly Ile Val Thr Pro Met Phe Leu Ala Lys Ile
180 185 190
Arg Lys Tyr Ala Tyr Phe Thr Leu Leu Val Ile Ala Ala Leu Ile Thr
195 200 205
Pro Pro Glu Leu Leu Ser His Met Met Val Thr Val Pro Leu Leu Ile
210 215 220
Leu Tyr Glu Ile Ser Ile Leu Ile Ser Lys Ala Ala Tyr Arg Lys Ala
225 230 235 240
Gln Lys Ser Ser Ala Ala Asp Arg Asp Val Ser Ser Gly Gln
245 250

<210> 9
<211> 245
<212> PRT
<213> Bacillus subtilis

<400> 9
Met Asp Lys Lys Glu Thr His Leu Ile Gly His Leu Glu Glu Leu Arg
1 5 10 15
Arg Arg Ile Ile Val Thr Leu Ala Ala Phe Phe Leu Phe Leu Ile Thr
20 25 30
Ala Phe Leu Phe Val Gln Asp Ile Tyr Asp Trp Leu Ile Arg Asp Leu
35 40 45
Asp Gly Lys Leu Ala Val Leu Gly Pro Ser Glu Ile Leu Trp Val Tyr

| | | | | | |
|---|-----|-----|-----|-----|-----|
| 50 | | 55 | | 60 | |
| Met Met Leu Ser Gly Ile Cys Ala Ile Ala Ala Ser Ile Pro Val Ala | | | | | |
| 65 | | 70 | | 75 | 80 |
| Ala Tyr Gln Leu Trp Arg Phe Val Ala Pro Ala Leu Thr Lys Thr Glu | | | | | |
| | 85 | | 90 | | 95 |
| Arg Lys Val Thr Ile Met Tyr Ile Met Tyr Ile Pro Gly Leu Phe Ala | | | | | |
| | 100 | | 105 | | 110 |
| Leu Phe Leu Ala Gly Ile Ser Phe Gly Tyr Phe Val Leu Phe Pro Ile | | | | | |
| | 115 | | 120 | | 125 |
| Val Leu Ser Phe Leu Thr His Leu Ser Ser Gly His Phe Glu Thr Met | | | | | |
| | 130 | | 135 | | 140 |
| Phe Thr Ala Asp Arg Tyr Phe Arg Phe Met Val Asn Leu Ser Leu Pro | | | | | |
| 145 | | 150 | | 155 | 160 |
| Phe Gly Phe Leu Phe Glu Met Pro Leu Val Val Met Phe Leu Thr Arg | | | | | |
| | 165 | | 170 | | 175 |
| Leu Gly Ile Leu Asn Pro Tyr Arg Leu Ala Lys Ala Arg Lys Leu Ser | | | | | |
| | 180 | | 185 | | 190 |
| Tyr Phe Leu Leu Ile Val Val Ser Ile Leu Ile Thr Pro Pro Asp Phe | | | | | |
| | 195 | | 200 | | 205 |
| Ile Ser Asp Phe Leu Val Met Ile Pro Leu Leu Val Leu Phe Glu Val | | | | | |
| | 210 | | 215 | | 220 |
| Ser Val Thr Leu Ser Ala Phe Val Tyr Lys Lys Arg Met Arg Glu Glu | | | | | |
| 225 | | 230 | | 235 | 240 |
| Thr Ala Ala Ala Ala | | | | | |
| | 245 | | | | |

<210> 10

<211> 63

<212> PRT

<213> Bacillus alcalophilus

<400> 10

| | |
|---|----|
| Met Gly Gly Leu Ser Val Gly Ser Val Val Leu Ile Ala Leu Val Ala | |
| 1 | 5 |
| | 10 |
| Leu Leu Ile Phe Gly Pro Lys Lys Leu Pro Glu Leu Gly Lys Ala Ala | |
| | 20 |
| | 25 |
| | 30 |
| Gly Ser Thr Leu Arg Glu Phe Lys Asn Ala Thr Lys Gly Leu Ala Asp | |
| | 35 |
| | 40 |
| | 45 |
| Asp Asp Asp Asp Thr Lys Ser Thr Asn Val Gln Lys Glu Lys Ala | |
| 50 | 55 |
| | 60 |

<210> 11

<211> 272

<212> PRT

<213> Bacillus alcalophilus

<400> 11

| | |
|---|----|
| Met Thr Met Met Thr Pro Asn Gln Gln Thr Ser Lys Lys Lys Lys Arg | |
| 1 | 5 |
| | 10 |
| | 15 |
| Lys Gly Arg Lys Gly Arg Val Pro Met Gln Asp Met Ser Ile Met Asp | |
| | 20 |
| | 25 |
| | 30 |
| His Ala Glu Glu Leu Arg Arg Arg Ile Phe Val Val Leu Ala Phe Phe | |
| | 35 |
| | 40 |
| | 45 |
| Ile Val Ala Leu Ile Gly Gly Phe Phe Leu Ala Val Pro Val Ile Thr | |
| | 50 |
| | 55 |
| | 60 |
| Phe Leu Gln Asn Ser Pro Gln Ala Ala Asp Met Pro Phe Asn Ala Phe | |
| 65 | 70 |
| | 75 |
| | 80 |

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Arg | Leu | Thr | Asp | Pro | Leu | Arg | Val | Tyr | Met | Asn | Phe | Ala | Val | Ile | Thr |
| | | | | 85 | | | | | 90 | | | | | 95 | |
| Ala | Leu | Val | Leu | Ile | Ile | Pro | Val | Ile | Leu | Tyr | Gln | Leu | Trp | Ala | Phe |
| | | | 100 | | | | | 105 | | | | | 110 | | |
| Val | Ser | Pro | Gly | Leu | Lys | Glu | Asn | Glu | Gln | Lys | Ala | Thr | Leu | Ala | Tyr |
| | | 115 | | | | | 120 | | | | | 125 | | | |
| Ile | Pro | Ile | Ala | Phe | Leu | Leu | Phe | Leu | Ala | Gly | Ile | Ala | Phe | Ser | Tyr |
| | 130 | | | | | 135 | | | | | 140 | | | | |
| Phe | Ile | Leu | Leu | Pro | Phe | Val | Ile | Ser | Phe | Met | Gly | Gln | Met | Ala | Asp |
| 145 | | | | | 150 | | | | | 155 | | | | | 160 |
| Arg | Leu | Glu | Ile | Asn | Glu | Met | Tyr | Gly | Ile | Asn | Glu | Tyr | Phe | Ser | Phe |
| | | | | 165 | | | | | 170 | | | | | 175 | |
| Leu | Phe | Gln | Leu | Thr | Ile | Pro | Phe | Gly | Leu | Leu | Phe | Gln | Leu | Pro | Val |
| | | | 180 | | | | | 185 | | | | | 190 | | |
| Val | Val | Met | Phe | Leu | Thr | Arg | Leu | Gly | Val | Val | Thr | Pro | Thr | Phe | Leu |
| | | 195 | | | | | 200 | | | | | 205 | | | |
| Arg | Lys | Ile | Arg | Lys | Tyr | Ala | Tyr | Phe | Ala | Leu | Leu | Val | Ile | Ala | Gly |
| | 210 | | | | | 215 | | | | | | 220 | | | |
| Ile | Ile | Thr | Pro | Pro | Glu | Leu | Thr | Ser | His | Leu | Phe | Val | Thr | Val | Pro |
| 225 | | | | | 230 | | | | | 235 | | | | | 240 |
| Met | Leu | Ile | Leu | Tyr | Glu | Ile | Ser | Ile | Thr | Ile | Ser | Ala | Ile | Thr | Tyr |
| | | | | 245 | | | | | 250 | | | | | 255 | |
| Arg | Lys | Tyr | His | Gly | Thr | Thr | Asp | His | Asn | Gly | Gln | Glu | Ser | Ala | Lys |
| | | | 260 | | | | | 265 | | | | | 270 | | |

<210> 12
 <211> 35
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> primer

<400> 12
 cccaagctta tgaaagggag ggcttttttg aatgg

35

<210> 13
 <211> 26
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> primer

<400> 13
 gcggatccaa agctgagcac gatcgg

26

<210> 14
 <211> 39
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> primer

<400> 14
 cccaagctta aaaagaaaga agatcagtaa gttaggatg

39

<210> 15
 <211> 25
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> primer

 <400> 15
 gcggatccaa gtcctgagaa atccg 25

 <210> 16
 <211> 21
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> primer

 <400> 16
 ggaattcgtg ggacggctac c 21

 <210> 17
 <211> 21
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> primer

 <400> 17
 cgggatccat catgggaagc g 21

 <210> 18
 <211> 26
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> primer

 <400> 18
 ggggtaccgg aaaacgcttg atcagg 26

 <210> 19
 <211> 22
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> primer

 <400> 19
 cgggatcctt tgggcgatag cc 22

 <210> 20

<211> 42
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> primer

 <400> 20
 gaggatccat gaggagagag gggatcttga atggcatacg ac 42

 <210> 21
 <211> 27
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> primer

 <400> 21
 cgatcctgca ggacctcatc ggattgc 27

 <210> 22
 <211> 27
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> primer

 <400> 22
 gtaggatccg cgcctaactt ctcaagc 27

 <210> 23
 <211> 25
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> primer

 <400> 23
 atagaattca aaaaggaaga gtatg 25

 <210> 24
 <211> 24
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> primer

 <400> 24
 ctggggatcc aaaaacagga aggc 24

 <210> 25
 <211> 35
 <212> DNA

<213> Artificial Sequence

<220>
<223> primer

<400> 25
gagaagggtcg acgcagcatt tacttcaaag gcccc 35

<210> 26
<211> 26
<212> DNA
<213> Artificial Sequence

<220>
<223> primer

<400> 26
accgggtcga ccgtcgtttt acaacg 26

<210> 27
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> primer

<400> 27
gggaattcat ggcctgcccg gtt 23

<210> 28
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> primer

<400> 28
caaggatccc gaattaagga gtgg 24

<210> 29
<211> 27
<212> DNA
<213> Artificial Sequence

<220>
<223> primer

<400> 29
ggtctgcagc tgcactaagc ggccgcc 27